Assessment of potential biomarkers in acetaminophen or thioacetamide-induced hepatic toxicity by siRNA

Jin Seok Kang[1], Young Na Yum[2] and Sue Nie Park[2]

[1]*Department of Biomedical Laboratory Science, Namseoul University*

[2]*Department of Toxicological Researches, National Institute of Toxicological Research, Korea Food and Drug Administration*

To compare between *in vivo* and *in vitro* profiles and to assess the feasibility between two systems, we have investigated global gene expression from both mouse liver and mouse hepatic cell line treated with hepatotoxicants, acetaminophen (APAP) and thioacetamide (TAA), respectively. By analyses of gene expression profiles, we picked up several down-regulated genes such as cytochrome P450 family 51 (Cyp51), sulfotransferase family cytosolic 1C member 2 (Sult1c2), 3-hydroxy-3-methylglutaryl-Coenzyme A synthase 1 (Hmgcs1), and several up-regulated genes such as growth arrest and DNA-damage-inducible 45 alpha (Gadd45a), transformation related protein 53 inducible nuclear protein 1 (Trp53inp1), zinc finger protein 688 (Zfp688) by APAP treatment. And we picked up several down-regulated genes such as aquaporin 8 (Aqp8), glutathione peroxidase 1 (Gpx1), succinate-CoA ligase, GDP-forming, alpha subunit (Suclg1), and several up-regulated genes such as chemokine (C-C motif) ligand 2 (Ccl2), DnaJ (Hsp40) homolog subfamily C member 5 (Dnajc5), tumor protein D52 (Tpd52). For validation of gene function, synthesized small interfering RNAs (siRNAs) for these genes were transfected in mouse hepatic cell line, BNL CL.2, and cell viability and RNA expression level were investigated. siRNA transfection rate was in proportion to its concentration (up to 100 nM) and was the highest for 48 h. We found siRNA transfection of these genes induced down-regulation of respective mRNA expression and decreased cell viability. Furthermore, short hairpin RNA transfection also reduced level of RNA expression. siRNA transfection for Cyp51, Aqp8 and others induced morphological alterations such as membrane thickening and nuclear condensation. Taken together, siRNA transfection of these genes induced alterations of cell viability and morphology as well as an effective inhibition of respective mRNA, and it suggests that these genes could be associated with APAP or TAA-induced toxicity. Furthermore, these genes may be used in the investigation of hepatotoxicity for better understanding of its mechanism.

Key words: acetaminophen, thioacetamide, toxicogenomics, hepatotixicty, siRNA